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EXPRESSION OF HUMAN NAT2 ACETYLTRANSFERASE OR BACTERIAL *O*-ACETYLTRANSFERASE IN CHO CELLS ENGINEERED TO STUDY THE GENOTOXICITY OF HETEROCYCLIC AMINES R.W. Wu, J.D. Tucker, K.J. Sorensen, L.H. Thompson, and J.S. Felton. Biology & Biotechnology Research Program, Lawrence Livermore Natl. Lab., Livermore CA, 94551.

To study heterocyclic amines produced in cooked food, we earlier developed the CHO UV5P3 cell line that expresses cytochrome P450IA2 and lacks nucleotide excision repair. The UV5P3 cells are ~50-fold more sensitive to PhIP than IQ with respect to cytotoxicity and mutation induction at the *aprt* locus. To test the hypothesis that the important missing activity in our CHO system for IQ genotoxicity was acetyltransferase, we transfected the UV5P3 cells with cDNA plasmids of either the human NAT2 acetyltransferase or a bacterial *O*-acetyltransferase used by M. Watanabe (kindly provided by Drs. T. Nohmi and T. Deguchi). Functionally transformed clones were determined by differential cytotoxicity assay using IQ, and confirmed by measuring the enzyme activity with isoniazid as substrate. Two clones designated 5P3NAT2 and 5P3YG (expressing human and bacterial transferases, respectively) were studied further. Both cell lines were sensitive to killing by IQ at concentrations as low as 0.004 µg/ml. Based on the 50% killing dose (D₅₀), the acetyltransferase expressing lines showed ~1000-fold increase in sensitivity over the parental UV5P3 cell line. The same dramatic change in sensitivity was also seen in mutation response at the *aprt* locus, and with chromosome aberrations and sister chromatid exchanges. In contrast, when these lines were exposed to PhIP, they showed cytotoxicity similar to that of the parental line UV5P3. These results suggest that PhIP does not require acetyltransferase for metabolic activation leading to genotoxicity in these cells. To study the role of DNA repair, the 5P3NAT2 cell line is being used to isolated repair-proficient revertants. These new lines constitute a sensitive cell system for assessing genotoxicity of compounds requiring metabolic activation by both P450IA2 and acetyltransferase, as well as studying the molecular processes by which DNA damage can lead to mutation. (This work was performed under the auspices of the U.S. DOE by LLNL under W-7405-Eng-48 and supported by NCI grant CA55861)